

**Section II. (Amendments to the Claims)**

Please cancel the non-elected claims, 142-187, 213-234, 242, 243, 250 and 251, with reservation of the right to file a divisional application directed to the subject matter thereof during the pendency of the present application, or a further divisional or continuing application based on and claiming priority of the present application.

Add new claims 262-280, and amend claims 190, 182, 212, 239 and 261, as set out below in the listing of claims 1-280 of the application.

1-187. (Cancelled).

188. (Previously presented) A method for culturing cells and producing a product of interest comprising:

- (a) providing a cell culture device comprising:
  - (i) a tangential flow device comprising:
    - (1) a housing enclosing an interior volume;
    - (2) one or more membranes separating the interior volume of the housing into a first set of one or more chambers and second set of one or more chambers;
    - (3) at least one inlet port and at least one outlet port in fluid communication with the first set of one or more chambers; and
    - (4) at least one port in fluid communication with the second set of one or more chambers;
  - (b) circulating a first culture medium fluid through the first set of one or more chambers of the tangential flow device;
  - (c) generating a permeate of the first culture medium fluid as a second fluid in the second set of one or more chambers of the tangential flow device;
  - (d) culturing cells to produce the product of interest; and
  - (e) collecting the first culture medium fluid and/or second fluid that contains the product of interest.

189. (Previously presented) The method of claim 188, wherein direction of said first culture medium fluid in said first set of one or more chambers of the tangential flow device is alternated by a switchable flow control device.
190. (Currently amended) The method of claim 189, wherein the switchable flow control device is selected from the group consisting of (i) reversible pump, (ii) 4-way valve, and (iii) a series of valves and (iv) reservoirs.
191. (Previously presented) The method of claim 188, wherein flow direction of said second fluid in said second set of one or more chambers is alternated by a switchable flow control device.
192. (Currently amended) The method of claim 191, wherein said switchable flow control device is selected from the group consisting of (i) reversible pump, (ii) 4-way valve, and (iii) a series of valves and (iv) reservoirs.
193. (Previously presented) The method of claim 188, further comprising the steps of monitoring the components and/or conditions of the first culture medium fluid and/or second fluid, and adding one or more substances selected from the group consisting of oxygen, carbon dioxide, serum, glucose, nutrients, and fresh culture medium into said first culture medium fluid and/or second fluid.
194. (Previously presented) The method of claim 193, wherein the cell culture device comprises multiple reservoirs for storing said one or more substances and adding same into said first culture medium fluid and/or second fluid.
195. (Previously presented) The method of claim 188, further comprising the step of treating the collected first culture medium fluid and/or second fluid that contains the product of interest, by a process selected from the group consisting of concentrating, separating, and dialyzing.
196. (Previously presented) The method of claim 195, wherein a filter device is used for treatment of the collected first culture medium fluid and/or second fluid containing the product of interest.
197. (Previously presented) The method of claim 196, wherein said filter device comprises a microporous filter and/or an ultrafilter.

198. (Previously presented) The method of claim 196, wherein said filter device comprises a tangential flow filtration element.
199. (Previously presented) The method of claim 188, further comprising the step of filtering the collected first culture medium fluid and/or second fluid with a microporous filter and/or an ultrafilter to yield a filtered fluid containing the product of interest.
200. (Previously presented) The method of 188, further comprising the step of filtering the collected first culture medium fluid and/or second fluid with a tangential flow filtration element to yield a filtered or concentrated fluid containing the product of interest.
201. (Previously presented) The method of claim 188, wherein said tangential flow device comprises one or more hollow fiber membranes that defines an intercapillary space and an extracapillary space.
202. (Previously presented) A method for culturing cells and producing a product of interest comprising:
- (a) providing a cell culture device comprising:
    - (i) a tangential flow device comprising:
      - (1) a housing enclosing an interior volume;
      - (2) one or more membranes separating the interior volume of the housing into a first set of one or more chambers and a second set of one or more chambers;
      - (3) at least one inlet port and at least one outlet port in fluid communication with the first set of one or more chambers; and
      - (4) at least one port in fluid communication with the second set of one or more chambers;
    - (b) flowing a culture medium fluid through the first set of one or more chambers of the tangential flow device;
    - (c) culturing cells to produce the product of interest;
    - (d) monitoring components and/or conditions of the culture medium fluid;
    - (e) adding one or more substances selected from the group consisting of oxygen, carbon dioxide, serum, glucose, nutrients, and fresh culture medium into said culture medium fluid; and
    - (f) collecting the culture medium fluid that contains the product of interest.

203. (Previously presented) The method of claim 202, wherein the cell culture device comprises multiple reservoirs for storing said one or more substances and adding same into said culture medium fluid.
204. (Previously presented) The method of claim 202, further comprising the step of treating the collected culture medium fluid that contains the product of interest, by a process selected from the group consisting of concentrating, separating, and dialyzing.
205. (Previously presented) The method of claim 204, wherein a filter device is used for treatment of the collected culture medium fluid containing the product of interest.
206. (Previously presented) The method of claim 205, wherein said filter device comprises a microporous filter and/or an ultrafilter.
207. (Previously presented) The method of claim 205, wherein said filter device comprises a tangential flow filtration element.
208. (Previously presented) The method of claim 202, further comprising the step of filtering the collected culture medium fluid with a microporous filter and/or an ultrafilter to yield a filtered fluid containing the product of interest.
209. (Previously presented) The method of 202, further comprising the step of filtering the collected culture medium fluid with a tangential flow filtration element to yield a filtered or concentrated fluid containing the product of interest.
210. (Previously presented) The method of claim 202, wherein said tangential flow device comprises one or more hollow fiber membranes that define an intercapillary space and an extracapillary space.
211. (Previously presented) The method of claim 202, wherein flow direction of said culture medium fluid in said first set of one or more chambers of the tangential flow device is alternated by a switchable flow control device.
212. (Currently amended) The method of claim 211, wherein said switchable flow control device is selected from the group consisting of (i) reversible pump, (ii) 4-way valve, and (iii) a series of valves and (iv) reservoirs.
- 213-234. (Canceled).

235. (Previously presented) The method of claim 188, wherein the culturing of cells occurs in the first set of one or more chambers of the tangential flow device.
236. (Previously presented) The method of claim 188, wherein the culturing of cells occurs in the second set of one or more chambers of the tangential flow device.
237. (Previously presented) A method of culturing cells and producing a product of interest, comprising (i) providing a multiplicity of passages bound by tangential flow membranes, including a first set of passages, a second set of passages, and a third set of passages, (ii) providing in one set of said first, second and third sets of passages cells which under culturing conditions produce the product of interest, (iii) flowing through another set of said first, second and third sets of passages a first medium essential to maintenance of said culturing conditions, (iv) flowing through a remaining set of said first, second and third sets of passages a second medium essential to maintenance of said culturing conditions, and (v) maintaining said culturing conditions while flowing said first medium through said another set of said first, second and third sets of passages, and flowing said second medium through said remaining set of said first, second and third sets of passages, whereby said cells in said one set of first, second and third sets of passages produce said product of interest.
238. (Previously presented) The method of claim 189, wherein flow direction of said second fluid in said second set of one or more chambers is alternated by an additional switchable flow control device.
239. (Currently amended) The method of claim 238, wherein said additional switchable flow control device is selected from the group consisting of (i) reversible pump, (ii) 4-way valve, and (iii) a series of valves and (iv) reservoirs.
240. (Previously presented) The method of claim 202, wherein the culturing of cells occurs in the first set of one or more chambers of the tangential flow device.
241. (Previously presented) The method of claim 202, wherein the culturing of cells occurs in the second set of one or more chambers of the tangential flow device.
- 242-243. (Canceled).

244. (Previously presented) The method of claim 193, wherein said one or more substances are filtered before they are added to the culture medium fluid with a microporous filter and/or an ultrafilter.
245. (Previously presented) The method of claim 203, wherein said one or more substances are filtered before they are added to the culture medium fluid with a microporous filter and/or an ultrafilter.
246. (Previously presented) The method of claim 188, wherein cells are cultured in the tangential flow device.
247. (Previously presented) The method of claim 188, wherein cells are cultured in suspension and/or in an anchored state.
248. (Previously presented) The method of claim 202, wherein cells are cultured in the tangential flow device.
249. (Previously presented) The method of claim 202, wherein cells are cultured in suspension and/or in an anchored state.
- 250-251. (Canceled).
252. (Previously presented) The method of claim 188, wherein the culture medium fluid is formulated for culturing cells selected from the group consisting of animal cells, human host cells, bacteria, mycobacteria, mycoplasma, and yeast.
253. (Previously presented) The method of claim 188, wherein the product of interest is selected from the group consisting of cells, bacteria, viruses, viral particles, intracellular products, extracellular products, IgG, immunoglobulins, hormones, proteins, amino acids, and macromolecules.
254. (Previously presented) The method of claim 202, wherein the culture medium fluid is formulated for culturing cells selected from the group consisting of animal cells, human host cells, bacteria, mycobacteria, mycoplasma, and yeast.

255. (Previously presented) The method of claim 202, wherein the product of interest is selected from the group consisting of cells, bacteria, viruses, viral particles, intracellular products, extracellular products, IgG, immunoglobulins, hormones, proteins, amino acids, and macromolecules.
256. (Previously presented) A method for culturing cells and producing a product of interest comprising: providing a flow circuit including a reservoir, a tangential flow membrane device and a reversible flow pump, wherein the reservoir, tangential flow membrane device and pump are connected in series in said flow circuit, with said tangential flow membrane device including membrane-separated first passages and second passages; culturing cells in said flow circuit; flowing material from the reservoir to said tangential flow membrane device to support said culturing; repetitively reversing flow in said first passages of said tangential flow membrane device by action of said pump; and generating a permeate including said product of interest, in said second passages of said tangential flow membrane device.
257. (Previously presented) The method according to claim 256, wherein said culturing cells in said flow circuit comprises culturing cells in first passages of said tangential flow membrane device.
258. (Previously presented) The method according to claim 256, wherein the cells cultured in said flow circuit comprise anchored cells.
259. (Previously presented) The method according to claim 256, wherein the cells cultured in said flow circuit comprise suspension cells.
260. (Previously presented) The method according to claim 256, further comprising use of at least one additional tangential flow device in said method.
261. (Currently amended) The method according to any one of claims 188 to 236 and 256 to 260, further comprising use of at least one additional tangential flow device in said method.
262. (New) The method according to any one of claims 189, 191, 211, and 238, wherein the switchable flow control device comprises a pump.

263. (New) The method according to any one of claims 189, 191, 211, and 238, wherein the switchable flow control device comprises a pump capable of reversing direction of the first culture medium fluid in the first set of one or more chambers of the tangential flow device.

264. (New) A method of culturing cells, comprising:

(a) providing a cell culture apparatus comprising:

- (1) at least one fluid reservoir;
- (2) a tangential flow membrane device in fluid communication with the at least one fluid reservoir and defining an intracapillary space and an extracapillary space;
- (3) at least one fluid exit port in fluid communication with the extracapillary space or the intracapillary space of the tangential flow membrane device; and
- (4) at least one pump in fluid communication with the tangential flow membrane device for circulating fluid through the intracapillary space of the tangential flow membrane device, wherein the pump is capable of alternatingly reversing direction of intracapillary space fluid flow through the tangential flow membrane device;

(b) circulating fluid through the intracapillary space of the tangential flow membrane device;

(c) alternatingly reversing direction of intracapillary space fluid flow through the tangential flow membrane device; and

(d) culturing cells in said cell culture apparatus.

265. (New) The method of claim 264, wherein the cell culture apparatus comprises at least one tangential flow membrane device.

266. (New) The method of claim 264, wherein said culturing comprises monitoring components and/or conditions in said cell culture apparatus affecting said cells, and adding one or more substances selected from the group consisting of oxygen, carbon dioxide, serum, glucose, nutrients, and fresh culture medium.

267. (New) The method of claim 266, wherein the cell culture apparatus comprises multiple reservoirs for storing said one or more substances and adding same.

268. (New) The method of claim 264, wherein the cell culture apparatus comprises a switchable flow control selected from the group consisting of a reversible pump, a four-way valve, a series of valves and reservoirs.



269. (New) The method of claim 264, further comprising filtering fluid from said cell culture apparatus to recover a product of interest.

270. (New) The method of claim 269, wherein said filtering comprises use of a microporous filter and/or an ultrafilter.

271. (New) The method of claim 264, further comprising collecting a culture medium fluid from said culturing and treating same.

272. (New). The method of claim 271, wherein said treating comprises a process selected from the group consisting of concentrating, separating, and dialyzing.

273. (New) A cell culture apparatus comprising:

- (1) at least one fluid reservoir;
- (2) a tangential flow membrane device in fluid communication with the at least one fluid reservoir and defining an intracapillary space and an extracapillary space;
- (3) at least one fluid exit port in fluid communication with the extracapillary space or the intracapillary space of the tangential flow membrane device; and
- (4) at least one pump in fluid communication with the tangential flow membrane device for circulating fluid through the intracapillary space of the tangential flow membrane device, wherein the pump is capable of alternatingly reversing direction of intracapillary space fluid flow through the tangential flow membrane device.

274. (New) The cell culture apparatus of claim 273, comprising at least one additional tangential flow membrane device.

275. (New) The cell culture apparatus of claim 273, further comprising a monitor adapted to monitor components and/or conditions affecting cell culture in said apparatus, and a source for one or more substances selected from the group consisting of oxygen, carbon dioxide, serum, glucose, nutrients, and fresh culture medium.

276. (New) The cell culture apparatus of claim 275, comprising multiple reservoirs for storing said one or more substances and adding same to fluid in said apparatus.

277. (New) The cell culture apparatus of claim 273, comprising a switchable flow control selected from the group consisting of a reversible pump, a four-way valve, a series of valves and reservoirs.

278. (New) The cell culture apparatus of claim 273, further comprising a filter adapted to filter fluid from said cell culture apparatus to recover a product of interest.

279. (New) The cell culture apparatus of claim 278, wherein said filter comprises a microporous filter and/or an ultrafilter.

280. (New) The cell culture apparatus of claim 273, further comprising a concentrator, separate or dialyzer arranged to treat culture medium fluid derived from cell culturing.